

THE SAN DIEGO-CONFERENCE

THE GENETIC REVOLUTION

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Hepatitis B DNA Detection

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POSTER TOPIC

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TEMP.

PERM.

COLORIMETRIC MICROPLATE HYBRIDIZATION ASSAYS FOR THE DETECTION OF HEPATITIS B VIRUS DNA IN SERUM. Christine L. Brakel, Enzo Diagnostics, Inc. An antibody-enhanced microplate hybridization assay (EMHA) was used for the direct detection of Hepatitis B Virus (HBV) DNA in serum. Eighteen of 60 (30%) Hepatitis B Surface Antigen (HBsAg) positive specimens contained HBV DNA while only 2 of 372 (0.5%) HBsAgnegative specimens were positive for HBV DNA. Forty-six specimens (37 HBsAg positive and 9 HBsAg negative) and 4 titration standards for the EMHA were tested independently for HBV DNA using the Abbott radiological Hepatitis B Viral DNA Hybridization test. Of 9 HBsAg-positive specimens that were positive for HBV DNA by the EMHA, 7 were positive by the Abbott The Abbott test identified 2 other HBV DNA-positive specimens (>3pg/ml DNA) as well as 2 specimens that contained equivocal amounts of HBV DNA (2 and 3.4pg/ml). Correlating the results of these two quantitative DNA hybridization tests indicated a detection limit for the EMHA of 15 to 20 pg of HB\! DNA.

In a separate study, the EMHA was again compared to the Abbott HBV DNA test and to the results of polymerase chain reaction (PCR) amplification of DNA isolated from serum. Those results confirmed a close correlation between the results of the EMHA and the Abbott test. As expected, both direct detection methods failed to detect several samples that contained HBV DNA when the nucleic acid was subjected to PCR amplification prior to assay in a standard Microplate Hybridization Assay. Unexpectedly, the results of PCR amplification indicated that in some specimens, there had been gene mutation and rearrangement in the Core antigen sequences of the Hepatitis B genome.

1988 ASM ANNUAL MEETING Miami Beach, Fla. 8–13 May 1988

Poster

Official Abstract Form

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Direct Detection of HSV Infection by DNA Hybridization, BRAKEL, C.L., POLLICE, M.A., and VICTOR, J. ENZO Biochem Inc., New York, N.Y.

The ColorGene™ DNA Hybridization Test for HSV Confirmation was applied to the direct detection of HSV-infected cells from herpes lesions. For this clinical trial 2 swabs from suspected herpes lesions were taken. The first swab specimen was used for culture isolation of HSV and material from the second swab was smeared onto glass microscope slides, fixed, and analyzed by in situ DNA hybridization. The results of this trial, carried out at 4 clinical virology laboratories, were 184 culture +/DNA +; 102 culture -/DNA -; 30 culture +/DNA -; and 24 culture -/DNA + (sensitivity = 86%, specificity = 81%). Of the 24 false positive specimens, 14 were from patients with a history of herpes infection, suggesting that the virus in these specimens had not grown in culture. On the average, specimens that tested as false negatives took longer to develop cytopathic effect in culture than specimens that tested true positive. The sensitivity of the test for vesicles, pustules, ulcers, and crusts was 92%, 87%, 82%, and 71%, respectively, indicating that direct testing is optimal for early as opposed to healing lesions, as is also true for isolation of virus in culture. Although the test does not distinguish HSV 1 from HSV 2, the test was equally sensitive for direct detection of both HSV types. The test is suitable for use in clinics and/or physicians' offices as the method requires no sophisticated equipment, is easy to interpret, and results can be observed with a simple light microscope. After fixation of the specimen (5-10 minutes), the test requires only 45-60 minutes until results can be read.

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